02-27-06 cgc

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Issued Patent No.	:	6,953,666
Issue Date	:	October 11, 2005
Appl. No.	:	09/831,123
Filing Date:	:	August 13, 2001
TC/A.U.	:	1641
Examiner	:	Lisa V. Cook
Applicant	:	Kinkade, Jr. et al.
For	:	Biomarkers for Oxidative Stress
Docket No.	:	68-97
Customer No.	:	23713

Certificate
MAR 01 2006
of Correction

Commissioner for Patents
Attention: Certificate of Corrections Branch
P.O. Box 1450
Alexandria, VA 22313-1450

CERTIFICATE OF MAILING
I hereby certify that this correspondence is being deposited with the United States Postal Service with sufficient postage for **Express Mail** in an envelope addressed to:
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February 24, 2006
Date

Cathy Nelson

EV 758 237 954 US
Express Mail Tracking Number

REQUEST FOR CERTIFICATE OF CORRECTION UNDER 37 C.F.R. 1.322

Sir:

Please issue a Certificate of Correction for U. S. Patent 6,953,666, as errors appear in the printed patent. Enclosed are two copies of Form PTO/SB/44 with the errors listed thereon. Also enclosed are copies of the specification pages referred to herein.

The printing errors appeared correctly in the application, as shown by the enclosed copies of the specification pages as originally filed.

The changes for "Other Publications" on the front page was listed correctly on the PTO Form 1449 filed with the Supplemental Information Disclosure Statement on January 3, 2003.

The error in column 2, line 41, appears correctly on page 3, line 8 of the application as originally filed.

The error in column 3, line 19, appears correctly on page 4, line 7 of the application as originally filed.

The errors in column 5, lines 15 and 16, appear correctly on page 6, line 27 of the application as originally filed.

The error in column 17, line 6, appears correctly on page 24, line 21 of the application as originally filed.

The error in column 24, line 35, appears correctly on page 35, line 23 of the application as originally filed.

The error in column 24, line 38, appears correctly on page 35, line 24 of the application as originally filed.

The error in column 27, line 60, appears correctly on page 40, line 11 of the application as originally filed.

The error in column 33, line 50, appears correctly in the Amendment filed January 28, 2004.

The error in column 35, line 18, appears correctly on page 52, line 3 of the application as originally filed.

All of the printing errors in claims 1, 20, and 32 appeared correctly in the Examiner's Amendment issued with the Notice of Allowance dated August 5, 2004.

The error in column 33, line 62, appeared correctly in the Examiner's Amendment issued with the Notice of Allowance dated August 5, 2004.

It is believed that the present submission does not require the payment of any fees. If this is incorrect however, please charge any required fee to Deposit Account No. 07-1969.

Respectfully submitted,

Susan K. Doughty
Reg. No. 43,595

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Attorney Docket No. 68-97
February 24, 2006

UNITED STATES PATENT AND TRADEMARK OFFICE

CERTIFICATE OF CORRECTION

PATENT NO. : **6,953,666 B2**

APPLICATION NO. : **09/831,123**

ISSUE DATE : **October 11, 2005**

INVENTOR(S) : **Kinkade, Jr. et al.**

It is certified that an error appears or errors appear in the above-identified patent and that said Letters Patent is hereby corrected as shown below:

Front Page:
Under the heading "Other Publications", first column, at the end of the "Ahmad" reference, last line, after 2, Jun. 1998, add --pages 87-92--.

In the Specification:
Column 2, line 41, replace "exposures" with --exposures.--.

Column 3, line 19, replace "mycloperoxidase" with --myeloperoxidase--.

Column 5, line 15, replace "ofexcess" with --of excess--.

Column 5, line 16, replace "cxidation" with --oxidation--.

Column 17, line 6, replace "c arbonic" with --carbonic--.

Column 24, line 35, replace "transfonnation" with --transformation--.

Column 24, line 38, replace "100-25 fold" with --100-fold--.

Column 27, line 60, replace "cystic" with --cysteic--.

Column 33, line 50, replace "allows" with --shows--.

Column 33, line 62, after "PTA-897.", insert --All restrictions upon public access to the deposit will be irrevocably removed upon the grant of a patent on this application and the deposit will be replaced if viable samples cannot be dispensed by the depository.--

Column 35, line 18, replace "(cystine." with --cystine.--.

In the Claims:
Column 38, Claim 1, line 31, replace "antibody" with --antibody produced by cell line K2.F1.6 (PTA-897)--.

Column 39, Claim 20, line 25, replace "antibody" with --antibody produced by cell line K2.F1.6 (PTA-897)--.

MAILING ADDRESS OF SENDER (Please do not use customer number below):

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UNITED STATES PATENT AND TRADEMARK OFFICE

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APPLICATION NO. : **09/831,123**

ISSUE DATE : **October 11, 2005**

INVENTOR(S) : **Kinkade, Jr. et al.**

Column 40, Claim 32, line 29, replace "antibody" with --antibody produced by cell line K2.F1.6 (PTA-897)--.

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ATTY DOCKET NO. 68-97	SERIAL NO.: 09/831,123	FILING DATE: Aug. 13, 2001
APPLICANT Kinkade et al.		GROUP: 1641

U.S. PATENT DOCUMENTS

Exmr Initial		Document Number	Date	Name	Class	Subclass	Filing Date if Appropriate

FOREIGN PATENT DOCUMENTS

		Document Number	Date	Country	Class	Subclass	Translation Yes/No
		96 04311 A	2/15/96	WO			
		97 11371 A	3/27/97	WO			
		98 12561 A	3/26/98	WO			

OTHER PRIOR ART *(including Author, Title, Date, Pertinent Pages, etc.)*

		Ahmad J. et al., "Detection of oxidative DNA damage by a monoclonal antibody: role of lysyl residues in antigen binding" *Immunology Letters*, Amsterdam, NL vol. 62, no. 2, June 1998 (1998-06), pages 87-92

EXAMINER **DATE CONSIDERED**

***EXAMINER:** Initial if reference considered, whether or not citation is in conformance with MPEP 609; Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant.

Reagents capable of oxidizing sulfur or selenium moieties in sulfur- or selenium-containing amino acids in proteins to sulfonic or selenocysteic acid moieties and sulfone or selenone moieties may be encountered directly in the environment (e.g., ozone), or may be generated endogenously, e.g., hypochlorous acid (HOCl) is generated by the myeloperoxidase (MPO)/hydrogen peroxide (H_2O_2)/chloride ion (Cl^-) system of activated phagocytic leukocytes during inflammation.

A number of exogenous sources of strong oxidants exist that are potentially important in chronic human exposures. These reportedly include ozone, radiation, chlorination processes that give rise to chloramines, oxides of nitrogen, iron and copper salts that promote oxidizing radical formation via Fenton chemistry, and normal dietary phenolic compounds (e.g., caffeic acid) that generate oxidants by redox cycling (Ames, B.N. et al. (1993), "Oxidants, antioxidants, and the degenerative diseases of aging," Proc. Natl. Acad. Sci. USA 90:7915-7922; Berlett, B.S. et al. (1996), "Comparison of the effects of ozone on the modification of amino acid residues in glutamine synthetase and bovine serum albumin," J. Biol. Chem. 271:4177-4182; Sies, H.,(ed.) (1991), *Oxidative Stress, Oxidants and Antioxidants*, New York: Academic Press; Stadtman, E.R. (1995), "Role of oxidized amino acids in protein breakdown and stability," Meth. Enzymol. 258:379-393; Thomas, E.L. et al. (1986), "Preparation and characterization of chloramines," Meth. Enzymol. 132:569-585; Fliss, H. and Ménard, M. (1994), "Rapid neutrophil accumulation and protein oxidation in irradiated rat lungs," J. Appl. Physiol. 77:2727-2733). Thus, interaction with a wide variety of environmental oxidants may also contribute to oxidative stress *in vivo* and the formation of oxidized sulfur or selenium moieties in sulfur- or selenium-containing amino acids, e.g., cysteic acid in proteins.

In addition, endogenous sources of strong oxidants include but are not limited to aerobic mitochondrial respiration, peroxisomes, and cytochrome P-450 enzymes (Ames, B.N. et al. (1993), "Oxidants, antioxidants, and the degenerative diseases of aging," Proc. Natl. Acad. Sci. USA 90:7915-7922; Sies, H. (ed.) (1991), *Oxidative Stress, Oxidants and Antioxidants*, New York: Academic Press).

Another major endogenous source of oxidative stress derives from the involvement of phagocytic leukocytes (neutrophils, monocytes/macrophages, eosinophils), which function in

defense against environmental and endogenous agents (Jesaitis, A.J. and Dratz, E.A. (eds.) (1992) *The Molecular Basis of Oxidative Damage by Leukocytes*, CRC Press, Boca Raton; Klebanoff, S.J. and Clark, R.A. (1978), *The Neutrophil: Function and Clinical Disorders*, North Holland, Amsterdam; Smith, J.A. (1994), "Neutrophils, host defense, and inflammation: a double-edged sword," J. Leukoc. Biol. 56:672-686). A major feature of this host defense function is a powerful oxygen-dependent, microbicidal, viricidal and tumoricidal system that utilizes two different peroxidases, myeloperoxidase (MPO) and eosinophilic peroxidase (EPO) (Henderson, W.R., Jr. (1991) "Eosinophil peroxidase: occurrence and biological function," in *Peroxidases in Chemistry and Biology*, Vol 1, Everse, J. et al. (eds.), CRC Press, Boca Raton, pp. 105-121; Klebanoff, S.J. (1992), "Oxygen metabolites from phagocytes," in *Inflammation. Basic Principles and Clinical Correlates*, J. I. Gallin et al. (eds.), Raven Press, NY, pp. 391-444). MPO is found only in the granules of neutrophils and monocytes/macrophages, and is biochemically distinct from EPO (Bainton, D.F. (1992), "Developmental biology of neutrophils and eosinophils," in *Inflammation: Basic Principles and Clinical Correlates*, 2nd ed., Gallin, J.L. et al. (eds.), Raven Press, NY, pp. 303-324; Henderson, W.R., Jr. (1991) "Eosinophil peroxidase: occurrence and biological function," in *Peroxidases in Chemistry and Biology*, Vol 1, Everse, J. et al. (eds.), CRC Press, Boca Raton, pp. 105-12; Nichols, B.A. and Bainton, D.F. (1973), "Differentiation of human monocytes in bone marrow and blood. Sequential formation of two granule populations," Lab. Invest. 29:27-40). Interaction of these cells with a variety of soluble and nonsoluble agonists leads to a respiratory burst (Gallin, J.I. et al. (eds.) (1992)*Inflammation: Basic Principles and Clinical Correlates*, Second Edition, Raven Press, Ltd., New York) and activation of an NADPH-dependent oxidase complex that generates large quantities of superoxide radical anion, a substantial portion of which dismutates to hydrogen peroxide (H_2O_2) (Segal, A.W. and Abo, A. (1993), "The biochemical basis of the NADPH oxidase of phagocytes," Trends Biochem. Sci. 18:43-47). In this process, cytoplasmic granules are mobilized and undergo secretion into both intracellular and extracellular spaces (Dahlgren, C. et al. (1989), "Localization of the luminol-dependent chemi-luminescence reaction in human granulocytes," J. Bioluminescence Chemiluminescence 4:263-266; Edwards, S.W. (1987), "Luminol- and lucigenin-dependent chemiluminescence of neutrophils: role of degranulation," J. Clin. Lab. Immunol. 22:35-39).

270:2906-2913). HOCl can also react with superoxide radical anion in a metal-ion independent Haber-Weiss type reaction to form hydroxyl radical ($^{\cdot}OH$) or with H_2O_2 to form singlet oxygen ($^1O_2$) (Candeias, L.P et al. (1993), "Free hydroxyl radicals are formed on reaction between the neutrophil derived species superoxide anion and hypochlorous acid," FEBS Lett. 333:151-153). Recent evidence suggests that Haber-Weiss chemistry also involves production of $^1O_2$ (Khan, A.U. and Kasha, M. (1994), "Singlet molecular oxygen in the Haber-Weiss reaction," Proc. Natl. Acad. Sci. USA 91:12365-12367).

Under acidic conditions in the presence of chloride ion, HOCl is in equilibrium with chlorine gas, and phagocytes have been shown to utilize this powerful oxidant at sites of inflammation and vascular disease (Hazen, S.L. et al. (1996), "Human neutrophils employ chlorine gas as an oxidant during phagocytosis," J. Clin. Invest. 98:1283-1289). Both phagocytic and endothelial cells produce nitric oxide (NO) and reaction of NO with superoxide leads to formation of peroxynitrite (Halliwell, B. (1996), "Antioxidants in human health and disease," Annu. Rev. Nutr. 16:33-50), a strong oxidant with properties similar to hydroxyl radical, which itself, can be formed by homolysis of peroxynitrite (Floris, R. et al. (1993), "Interaction of myeloperoxidase with peroxynitrite. A comparison with lactoperoxidase, horseradish peroxidase and catalase," Eur. J. Biochem. 215:767-775). In principle, all of these reactive oxygen species (ROS) and reactive non-oxygen species are strong enough to oxidize the sulfur or selenium moieties of sulfur- or selenium-containing amino acid residues in proteins to cysteic acid (cysteine sulfonic acid) or selenocysteic acid and to sulfone or selenone moieties such as methionine sulfone or selenone.

In vitro studies with model compounds show that HOCl reacts at least 100 times faster with thiols compared to primary amines (Folkes, L.K. et al. (1995), "Kinetics and mechanisms of hypochlorous acid reactions," Arch. Biochem. Biophys. 323:120-126; Winterbourn, C.C. (1985), "Comparative reactivities of various biological compounds with myeloperoxidase-hydrogen peroxide-chloride, and similarity of the oxidant to hypochlorite," Biochim. Biophys. Acta 840:204-210). In the presence of excess thiol, HOCl oxidation leads to disulfide formation (Silverstein, R.M. and Hager, L.P. (1974), "The chloroperoxidase - catalyzed oxidation of thiols and disulfides to sulfenyl chlorides," Biochemistry 13:5069-5073). However, in the absence of excess thiol, (a condition that exists *in vivo* at sites of inflammation) (Fliss, H. and Ménard, M.

parameters) can then be compared to individuals with pathophysiological conditions that are related to inflammation and oxidative stress, e.g., coronary artery disease, diabetes, etc.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 is a chromatographic profile showing the HPLC separation, identification and quantification of the D and L isomers of cysteic acid (CA), L-cysteine sulfinic acid (L-CSA) and D and L-aspartic acid (ASP) and the internal standard D-2 amino-3-phosphopropionic acid (APPA).

Figure 2 is a graph of cysteic acid (picomoles) versus LDL apoprotein (micrograms) showing a least squares linear regression plot of data from the hydrolysis of 3 concentrations of 2 different delipidated human LDL samples.

Figure 3 is a graph of cysteic acid (picomoles)/protein (micrograms) versus sample number, samples 1-15 being from healthy humans, samples 16 and 17 from human patients with renal disease, samples 18-20 from human patients with unspecified diagnoses, samples 21-25 from human patients with coronary artery disease (CAD).

Figure 4 is a graph of cysteic acid (picomoles)/protein (micrograms) versus FPLC fraction number, protein (milligram/100 microliters) versus FPLC fraction number, and cholesterol (milligram/milliliter) versus FPLC fraction number.

Figure 5A is SDS-PAGE of oxidized and unoxidized proteins using murine antiserum raised against the oxidized A-chain of bovine insulin. Each lane contained 0.5 μg of the following proteins: (1) prestained M_r markers; (2) unoxidized glyceraldehyde-3-phosphate dehydrogenase (GAPDH); (3) H_2O_2-oxidized GAPDH; (4) carbonic anhydrase; (5) oxidized A-chain of bovine insulin conjugated to ovalbumin, oxA-OVA; (6) OVA; (7) M_r markers; (8) bovine insulin; (9) H_2O_2-oxidized BSA; (10) unoxidized BSA.

Figure 5B is Western blotting of oxidized and unoxidized proteins using murine antiserum raised against the oxidized A-chain of bovine insulin. The lanes correspond to those in Figure 5A.

29) has been isolated from this protein (Pohl, J. et al. (1982), "Identification of the active site cysteine and of the disulfide bonds in the N-terminal part of the molecular of bovine spleen cathepsin B," FEBS Lett. 142:23-26), and the content of cysteic acid (based on yield of peptide) was estimated to be 5% of the Cys-29 residue. As a negative control, analyzed human MBP which contains no cysteine/cystine was analyzed. The results of these analyses are shown in Table 1. The finding of 4.5% cysteic acid in Cat B agreed well with the estimated value of 5%. MBP, LZ and RNase showed no significant amount of cysteic acid over blank values. BSA and Oval both showed significant amounts of cysteic acid over background levels, and these results were reproducible (less than 10% variation) when repeated with different samples.

TABLE 1

Protein	MW $x10^{-3}$	Thiol (SH) mole/mole protein	Disulfide (S-S) mole/mole protein	Cysteic Acid mole/mole unoxidized protein	Cysteic Acid mole/mole HOCl-oxidized protein
BSA	66.4	1	17	0.017	0.8
Oval	42.7	4	1	0.008	1.6
CatB	27.5	2	7	0.045	N.D.
MBP	18.4	0	0	0	0
LZ	14.3	0	4	0	0.36
RNase	13.7	0	4	0	0.60

N.D. = not determined

EXAMPLE 3: Quantification of Cysteic Acid in HOCl-Oxidized Proteins

Oxidation Using Reagent HOCl. Proteins were subjected to *in vitro* HOCl oxidation under conditions consistent with those occurring at *in vivo* sites of inflammation: molar ratio of HOCl:Protein = 800:1 (Hazell, L.J. and Stocker, R. (1993), "Oxidation of low-density lipoprotein with hypochlorite causes transformation of the lipoprotein into a high-uptake form for macrophages," Biochem. J. 290:165-172). The reaction was stopped by quenching with a 100-fold molar excess of methionine and allowed to remain at room temperature for 1-2 h to ensure that no protein-associated chloramines remained (Hazell, L.J. et al. (1994), "Oxidation of low-density lipoprotein by hypochlorite causes aggregation that is mediated by modification of lysine residues rather than lipid oxidation," Biochem. J. 302:297-304). The samples were dialyzed to

amino acids are performed using the chemical method of the present invention described above or other methods known in the art or methods readily adapted from those known in the art. Reduced and carboxymethylated preparations of the same proteins are used as negative controls. Candidate mAbs are further characterized for IgG isotype and reactivity with a larger panel of performic acid-oxidized proteins. mAbs are purified from ammonium sulphate-precipitated culture supernatants by affinity chromatography with protein A-Sepharose. In addition, the fine-specificity of the candidate monoclonal antibodies are tested with the following related chemical moieties by competition in ELISAs: cysteic acid and homocysteic acid, taurine, β-alanine, phosphoserine, phosphothreonine and phosphotyrosein. For larger quantities of mAbs, ascites are generated in mice.

Alternatively, groups of mice can be immunized with cysteic acid conjugated to Keyhole Limpet Hemocyanin (KLH), a procedure that was used in producing monoclonal antibodies to phosphoserine and phosphothreonine (Abu-Lawi, K.I. and Sultzer, B.M.(1995), "Induction of serine and threonine protein phosphorylation by endotoxin-associated protein in murine resident peritoneal macrophages," Infect. Immun. 63:498-502; Hasegawa, M. et al. (1996), "Characterization of mAb AP422, a novel phosphorylation-dependent monoclonal antibody against tau protein," FEBS Lett. 384:25-30). Preparation of the cysteic acid-KLH conjugate involves standard protein chemistry as will be understood by those of ordinary skill in the art.

The initial approach here was to immunize mice with the oxidized A-chain of bovine insulin (a 21-mer obtained by performic acid oxidation), an immunogen of known structure that contains four cysteic acid residues. An HPLC procedure for screening sera against both oxidized proteins and unoxidized, control proteins that can detect as little as four picomoles cysteic acid per microgram oxidized protein was developed. These proteins were probed with mouse antisera or pre-non-immune sera, followed by a goat antimouse alkaline phosphatase conjugated antibody, and the signal is quantified and analyzed using an automated, computer-driven microplate reader (ELISA assay). Screening mouse sera for production of a significant antibody titer avoids beginning the time and labor-intensive procedures of cell fusions, selection and screening for positive hybridomas if the animals are not responding positively to the immunogen. The development of the specific procedures and the data reported in this section were obtained using polyclonal mouse antisera.

amount of partially oxidized cystine in the oxidized CAP-37 preparation could explain the small reactivity that was observed (Table 4). It is clear that the mAb is recognizing an oxidation-specific epitope in the protein that involves cysteine and/or cystine.

EXAMPLE 9. Human sera plasma testing for endogenous antibody to oxidized protein

The ELISA assay developed to screen for mAb was used to look for the presence of endogenous antibody to oxidatively damaged protein (ODP) in the plasma of normal individuals and patients with coronary artery disease (CAD) and renal disease. 96-well microplates were coated with PAoxOVA and blocked with gelatin. Diluted plasma was added to the plates and the presence of endogenous antibody was detected and quantified by addition of a goat anti-human IgG(Fc)-alkaline phosphatase conjugate. As described above, the kinetic data are reported as Vmax values (milliAbsorbance(OD) units/minute at 405 nm). In most cases, these clinical data represent values from a single well.

Children. Two sets of serum samples (110 total) were obtained from children ages 12-13 years. The first set (samples 1-89) was from a recently discovered aboriginal tribe in the Phillipines, and the ages ranged from 13-16 years. Many of these children have high levels of Lpa, a risk factor for development of CAD. The second set (samples 90-110) was from a group of American 12-13 year-olds who have been determined to be at risk for developing CAD (e.g. tendency toward obesity and/or have parents who are diabetic or obese or have other risk factors for CAD). The results of this study using plasma diluted 1:200 are shown in Fig. 12A and Fig. 12B. Two points can be made. A number of these children have rather high levels of endogenous antibody to oxidatively damaged protein as measured by reactivity against PAoxOVA. Second, the younger group of American children (ages 12-13) appear to have lower levels of endogenous antibody against oxidatively damaged protein (ODP) than the somewhat older Philippine group (ages 13-16): American group (n = 21): mean +/- S.D. = 8.6 +/- 7.3; median = 6.3; Philippine group (n=89): mean +/- S.D. = 19.0 +/- 16.8; median = 12.7. Further testing of a considerably larger number of subjects will be required to determine the range of values for "normals" and to determine if these levels increase with age, and if there are differences between males and females.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Appl. No.	:	09/831,123	Confirmation No. 8722
Applicant	:	Kinkade et al.	
Filed	:	August 13, 2001	
TC/A.U.	:	1641	
Examiner	:	Cook, Lisa V.	
For	:	BIOMARKERS FOR OXIDATIVE STRESS	
Docket No.	:	68-97	
Customer No.	:	23713	

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CERTIFICATE OF MAILING
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PO Box 1450, Alexandria, VA 22313-1450

January 28, 2004
Date

Cathy Nelson

EV 412171595 US
Express Mail Tracking Number

RESPONSE TO OFFICE ACTION

Sir:

In response to the Office Action of July 29, 2003, please amend the above-identified application as follows:

Amendments to the Specification begin on page 2 of this paper.

Amendments to the Claims are reflected in the listing of claims which begins on page 4 of this paper.

Remarks/Arguments begin on page 16 of this paper.

Amendments to the Specification:

Please replace the Cross Reference to Related Applications Section (paragraph 1 on page 1) with the following amended paragraph:

This application is the National Stage of International Application No. PCT/US99/26133, filed November 5, 1999, which takes priority from United States provisional Patent Application No. 60/107,404, filed November 6, 1998.

Please replace paragraph 2 on page 48, lines 13-22, with the following amended paragraph:

Tables 3-5 show the activity expressed as Vmax values (milliAbsorbance(OD) units/minute at 405 nm) for 3 different clones of the mAb (K2.F1.1, K2.F1.3 and K2.F1.6) and a non-producing clone used as a negative control (K2.A12). Table 3 shows the results for wells coated with 1μg OVA-unoxCAP 37. Table 4 shows the results for wells coated with 1 μg OVA-oxCAP 37. Table 5 shows the results for wells coated with 1μg PAox OVA. No activity was seen using the unoxidized CAP-37 polypeptide conjugated to OVA (Table 3), but, surprisingly, very little activity was observed when the oxidized CAP-37-OVA conjugate was used in the screen (Table 4). Figure 11 shows the template of a 96-well plate used in obtaining the data of Tables 3-5. The cell line K2.F1.6 was deposited with the American Type Culture Collection (ATCC) 10801 University Blvd., Manassas, VA, 20110-2209 on October 29, 1999, and assigned Patent Deposit Number PTA-897.

EXAMINER'S AMENDMENT

1. An examiner's amendment to the record appears below. Should the changes and/or additions be unacceptable to applicant, an amendment may be filed as provided by 37 CFR 1.312. To ensure consideration of such an amendment, it MUST be submitted no later than the payment of the issue fee.

2. Authorization for this examiner's amendment was given in a telephone interview with Susan K. Doughty (Reg. No. 43,595) on 7/23/04.

I. Cancel claims 41-84 without prejudice or disclaimer.

II. In claim 1 line 2 after "an antibody" insert --produced by cell line K2.F1.6 (PTA-897)--.

III. In claim 23 line 2 after "an antibody" insert --produced by cell line K2.F1.6 (PTA-897)--.

IV. In claim 37 line 2 after "an antibody" insert --produced by cell line K2.F1.6 (PTA-897)--.

V. In the specification on page 48, paragraph 2, line 22, after "PTA-897." Insert --All restrictions upon public access to the deposit will be irrevocably removed upon the grant of a patent on this application and the deposit will be replaced if viable samples cannot be dispensed by the depository.--

3. **NO EXTENSIONS OF TIME ARE PERMITTED TO FILE CORRECTED OR FORMAL DRAWINGS, OR A SUBSTITUTE OATH OR DECLARATION**, notwithstanding any indication to the contrary in the attached Notice of Allowability (PTO-37).